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RESEARCH ARTICLE

Post-Mortem Diagnosis of a Rare Biphasic Benign Multicystic Peritoneal Mesothelioma in an Indian Domestic Fowl (Gallus Domesticus)

***B. Dhanush Krishna[1], Sherin B. Sarangom[2], N. Divakaran Nair[1], Syam K. Venugopal[2], V. Dhanesh[3]**

1 Department of Veterinary Pathology, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala, India – 680651

2 University Veterinary Hospital and TVCC, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala, India – 680651

3 University Poultry and Duck Farm College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala, India – 680651

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Corresponding Author

B. Dhanush Krishna

Abstract

Mesotheliomas are rare neoplasms originating from coelomic cavities. Environment pollution, viral or genetic factors have been implicated in the pathogenesis of this neoplasms. Although several cases are on record in man and animals, the veterinary medical literatures on mesotheliomas in birds are scarce especially those affecting poultry. Also, the biphasic pattern of mesothelioma is rare compared to other histological structural forms. In the present case, a seven month-old male bird of Gramasree breed was diagnosed with biphasic benign multi-cystic peritoneal mesothelioma (BMPM) during necropsy. This was a rare case of biphasic BMPM in an Indian Domestic fowl and hence required documentation.

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INTRODUCTION

Mesotheliomas are uncommon primary tumours originating from mesothelial cells lining the body cavities such as peritoneum, pleura, pericardium and tunica vaginalis (Barker, 1993), (Morini et al, 2006), (Vural et al, 2007), (Inai, 2008). The tumor was reported to have encountered in domestic, wild, companion and laboratory animals as wells as birds and man (Patnaik and Mohanty, 1970), (Barker 1993), (Jones et al., 1997), (Braun et al, 2004), (Wiedner et al, 2008), (Chua et al, 2009). Although mesotheliomas were described in domestic as well as birds of prey over the last century (Patnaik and Mohanty, 1970), (Cooper and Pugsley, 1984), reports of this tumour in birds, especially in domestic fowl, during the last two decades are relatively few in number. In humans, 70 percentage of the reported cases of mesothelioma account for pleural mesothelioma and the remaining 30 percent account for peritoneal mesothelioma (Chua et al, 2009). The veterinary literatures on peritoneal mesothelioma were also found to be scarce. Among the three histologic forms of mesothelioma, the biphasic and sarcomatoid forms are rare compared to the epitheliod mesothelioma (Inai, 2008), (Chua et al, 2009). Currently, exposure to asbestos, iron or silica has been incriminated in the development of mesotheliomas in addition to viral etiology and genetic factors (Barker, 1993), (Hillerdal, 1999), (Lopez, 2001), (Head et al, 2002), (Inai, 2008), (Chua et al, 2009). Also, the researches on proposed causes in the development of mesothelioma are underway. The clinical and pathologic findings of a rare biphasic benign multicystic peritoneal mesothelioma (BMPM) in an Indian domestic fowl with original breed name Gramasree are hence documented. The Gramasree is a poultry strain developed by the Centre for Advanced Studies in Poultry Science, College of Veterinary and Animal Sciences, Thrissur, Kerala, India, well suited for backyard poultry farming due to its foraging and scavenging nature with an average yield of 200 eggs per year (Sathiadhas et al, 2006).

Case Report

A 7 month-old male Indian domestic fowl (Gallus domesticus), Gramasree, bred and housed in the Kerala Veterinary and Animal Sciences University Poultry and Duck Farm, Mannuthy, Thrissur had the history of abdominal distension, anorexia, and dyspnoea for the previous week. The remaining flock of birds was reported to be apparently normal by the staff and did not show any simultaneous illness. The bird was regularly dewormed, vaccinated and was fed with formulated poultry feed supplemented with vitamins and minerals. A tentative diagnosis of non-specific respiratory infection was made and the bird was treated with antibiotics and anti-inflammatory drugs. But the bird succumbed before any diagnostic evaluation could be performed. A detailed necropsy of the carcass was performed in the Department of Veterinary Pathology, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala.

At necropsy, a massive, freely floating mass was noticed in the abdominal cavity that was found attached to the dorsal parietal peritoneum at the level of lumbo-sacrum and occupied much of the cavity (**Fig. 01**). The mass was found to have reddish white, soft, nodular and multi-cystic structure with occasional coalescence, covered by a transparent glistening capsule, containing about 250 milliliters of serosanguinous fluid and measured approximately 4cm × 4cm x 3 cm. It appeared as if floating over and into the interspaces of abdominal organs. Any signs of inflammatory reactions or exudations were not observed anywhere in the abdominal cavity. Gross evidence of any malignancy and involvement with abdominal organs like liver, spleen, gallbladder, kidneys, intestines, gizzard and proventriculus were not noticed. Tissue samples were obtained from major visceral organs as well as nodular multi-cystic lesions and were fixed in 10% neutral buffered formalin. The representative tissue samples were paraffin- embedded, sectioned at 4µm, and stained with Haematoxylin and Eosin (H&E). The sections were also subjected to special stains like Masson's trichrome, Periodic acid-Schiff reaction with and without diastase digestion and Alcian blue (pH 2.5) with and without hyaluronidase for histopathological confirmation.

Microscopically, there were cuboidal epithelial cells supported by a conspicuous stroma formed papilliform ridges in certain areas of the tumour. There were spindle cells, which varied in size and had oval nuclei and scanty cytoplasm. Scattered large oval to round epithelioid cells with peripheral nuclei, distinct cell borders, and abundant eosinophilic cytoplasm were also observed within the same tumour. Mitotic figures were rare, but tumour growth was extensive. Epithelioid neoplastic cells had nuclei with loosely distributed chromatin and generally contained a single nucleolus. Papillo-tubular structures of epithelial components with proliferating spindle cells were seen as a prominent feature in the majority of the tumour mass (**Fig. 03**). Neoplastic cells did not invade the underlying tissue. Collagen was present between the tumor masses. The mucins in the cytoplasm of the cells were positively stained with Alcian blue but did not react with Periodic acid-Schiff stain after diastase pretreatment (**Fig. 05**). Masson's trichrome stained sections revealed fine fibrils and broad bundles of collagenous stroma supporting and separating papillo-tubular structures of epithelial components (**Fig. 04**). The condition was diagnosed as biphasic BMPM of unknown etiology based on cellular morphology.

Discussion

Benign Multicystic Peritoneal Mesothelioma (BMPM) also known as multicystic peritoneal inclusion cyst is an uncommon neoplasm arising from the peritoneal mesothelium (Inman et al, 2000), (Safioleas et al, 2006). Although the origin of the tumour is known, the incidence, etiology, pathogenesis and the natural behaviour remain unclear. Early diagnosis of peritoneal mesothelioma is often difficult because the early symptoms mimic an ailment of the gastrointestinal tract or the respiratory system as in the present case. In most cases, the tumour remains silent in abdominal cavity and may invade several underlying organs and at times, even the retro peritoneum with malignant transformation. Therefore, diagnosis often occurs at an advanced stage when the neoplasm spreads throughout the peritoneal cavity (Inai, 2008), (Chua et al, 2009). These tumours are diagnosed incidentally at times during clinical examination or laparotomies and often during necropsy. Even though advanced imaging techniques might enable to demonstrate the lesion (Inman et al, 2000), (Safioleas et al, 2006), a differential diagnosis is difficult to be made from other cystic neoplastic or inflammatory lesions arising from these anatomical areas.

In humans and rats, three histological forms of mesothelioma are described viz., epitheliod, sarcomatoid or biphasic form (Jones et al, 1997), (Chua et al, 2009), (Ohnuma-Koyama et al, 2013). Epitheliod mesotheliomas are more common compared to the biphasic and sarcomatoid type (Head et al, 2002), (Inai, 2008), (Chua et al, 2009). The biphasic subtype shows a mixture of both epitheloid and sarcomatoid features characterized by moderate to severe cytological atypia. Besides these three major types, several subtypes have also been described viz., lymphohistiocytoid, desmoplastic, microcystic, clear cell, small cell and deciduoid (Shibuya et al, 1990), (Head et al, 2002), (Morini et al, 2006). In the present case, the tumour was categorized based on gross necropsy findings, histological pattern and staining characteristics. Histological biphasic appearance was observed differentiating into epithelioid cells lining the peritoneal surface and underlying spindle cells in varying proportions, as observed in many studies (Inman et al, 2000), (Inai, 2008), (Chua et al, 2009). Mitotic figures were frequent in some cases previously reported (Head et al, 2002). However, in this case mitotic figures were scarce, with less than one per high-power field.

From the clinical point of view, BMPM has to be differential diagnosed from adenocarcinomas originating from different visceral organs like intestine, gizzard and proventriculus. Grossly, no neoplastic lesions were found in these organs. Though the tumour in this bird did not show any involvement or invasion of surrounding visceral organs, the

harmful effects might have been mechanical due to the size and location of the tumour as previously reported (Patnaik and Mohanty, 1970). In female birds, adenocarcinomas involving ovaries also have to be ruled out (Barker, 1993), (Yamate et al, 2007). Ultrasonographic evaluation or a pre-operative fine needle aspiration biopsy of the cystic lesions may aid in differentially diagnosing the condition (Braun et al, 2004), (Safioleas et al, 2006). Adenocarcinomas are histologically characterized by tubular or acinar patterns where the spaces are lined with cuboidal to columnar epithelial cells (Battifora and Kopinski, 1985), (Kumar et al, 2004), (Yamate et al, 2007). Such growth patterns were not observed in the present case. Also, special staining has been performed in distinguishing mesothelioma from adenocarcinoma. Adenocarcinomas typically contain neutral mucins that stain with the Periodic acid–Schiff stain after diastase pretreatment. Mesotheliomas, on the other hand, secrete acid mucins (predominantly hyaluronic acid) that stain with alcian blue but not with Periodic acid–Schiff stain after diastase pretreatment (Battifora and Kopinski, 1985), (Kumar et al, 2004), (Yamate et al, 2007). Immunohistochemical evaluation and electron microscopic examination could be employed as they provide conclusive answers (Inai, 2008), (Chua et al, 2009).

From the available veterinary medical literatures, mesotheliomas were frequently encountered in cattle and dogs (McDonough et al, 1992), (Barker, 1993), (Head et al, 2002), (Braun et al, 2004), (Reggeti et al, 2005), (Vural et al, 2007). This tumour was also noticed in buffaloes, horses, cats, goats, sheeps, pigs, tiger, leopard, lion, macaque, hamsters, rabbits, rats and mice (Singh and Singh, 1970), (Carnine et al, 1977), (Raflo and Nuernberger, 1978), (Barker, 1993), (Cicala et al, 1993), (Cunningham and Dhillon, 1998), (Head et al, 2002), (Yener et al, 2002), (Kutsal et al, 2003), (Morini et al, 2006), (Yamate et al, 2007), (Wiedner et al, 2008), (Bollo et al, 2011), (Ohnuma-Koyama et al, 2013). Among birds, mesothelioma has been reported in Ferruginous hawk and fowl decades ago (Patnaik and Mohanty, 1970), (Cooper and Pugsley, 1984). Congenital mesotheliomas are encountered in calves, while in all other species the occurrence of this neoplasm positively correlates with age (Magnusson and Veit, 1987), (Head et al, 2002). Preponderance towards the male sex as in humans (Chua et al, 2009) was not noticed in animals and birds. In addition, mesothelioma have been experimentally induced by asbestos in rats, avian leukosis virus MC29 in chicken, papova virus SV40 infection in hamsters and rats and polyoma virus in swiss mice (Chabot et al, 1970), (Head et al, 2002), (Cicala et al, 1993). Even chrysotile asbestos and ferruginous bodies suggestive of asbestos exposure have been identified in the lung tissues of urban dogs with mesotheliomas (Glickman et al, 1983), (Harbinson and Godleski, 1983). Also, the development of pleural mesothelioma after exposure to asbestos is well documented in humans (Hillerdal, 1999), (Inai, 2008), (Chua et al, 2009). However this association has not been confirmed in animals. Moreover, the bird in the present report was not known to have been exposed environmentally to any inducing or infectious agents of mesothelioma.

Conclusion

A few cases of peritoneal mesothelioma have been reported in poultry. These tumours are much less common than their pleural counterparts. The present case of biphasic mesothelioma was found to be rare compared to the other mesotheliomas reported in birds. Obviously, for the evaluation and assessment of the natural behavior and clinical characteristics of this tumor, the domestic fowl can often be considered as a novel model. However, the investigations on determining the etiological factors behind this disease, other than the postulated and proven causes require strategic evaluation. Greater awareness of species susceptibilities also enables clinicians to make an earlier and accurate diagnosis, thus improving case prognosis. The present findings help both clinicians and pathologists to take a concerted effort to improve the accuracy in the diagnosis of various types of mesothelioma.

Fig. 1: Fowl with cauliflower-like growths embedded among multi-cystic fluid filled structures on the peritoneal cavity

Fig.2: The excised tumour mass revealed multiple, irregularly round and reddish white small nodular units

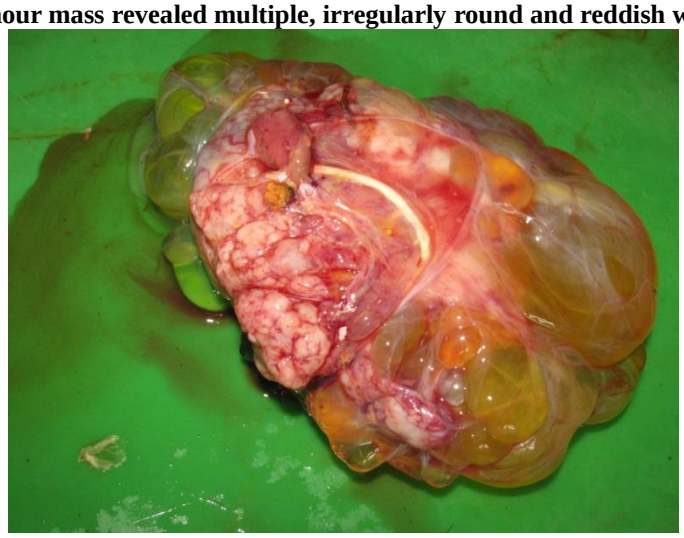

Fig.3: Microphotograph of biphasic mesothelioma (H&E stain, X400). Papillo-tubular structures of epithelial components were seen in between proliferating spindle cells

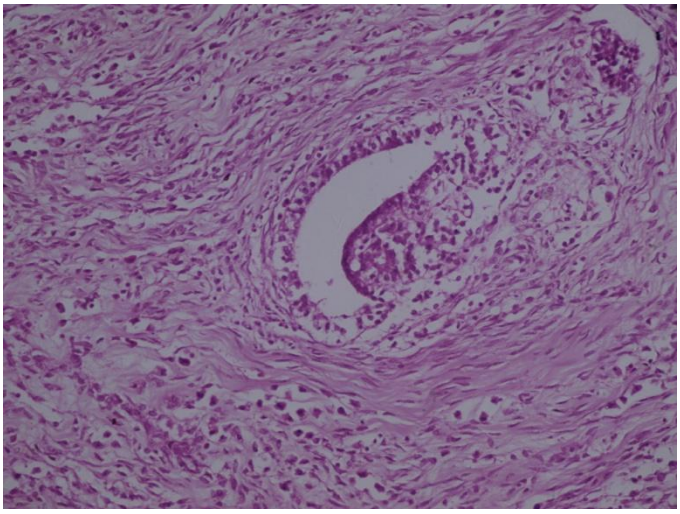

Fig.4: Microphotograph of biphasic mesothelioma (Masson's trichrome stain, X400). The epithelial and mesenchymal components were mixed within one tumour.

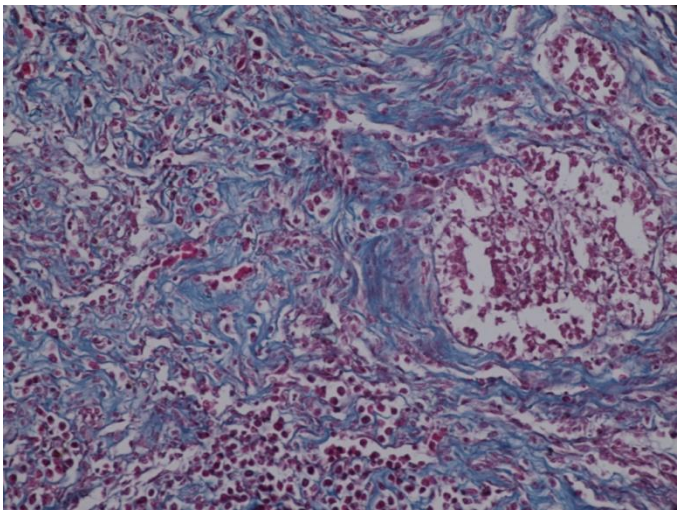

Fig.5: Microphotograph of biphasic mesothelioma (Alcian blue stain, X400). Acid mucins (predominantly hyaluronic acid) were demonstrated.